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# Effect of oxidation with coagulation and ceramic microfiltration pre-treatment on reverse osmosis for desalination of recycled wastewater

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## Abstract

Oxidation and coagulation before ceramic microfiltration (CMF) greatly increases membrane flux, but is unconventional for reverse osmosis (RO) pre-treatment. Impacts to RO and the wastewater recycling scheme operating CMF at high flux conditions is little understood. In this work, wastewater was treated with ozone or ultraviolet/hydrogen peroxide (UVH) oxidation, coagulation, then CMF, to explore RO membrane performance at bench scale. Sustainable high CMF fluxes were confirmed using coagulation with either ozone or UVH. Uniquely for ozone, dosing 13 mg-O<sub>3</sub>/L for 15 minutes greatly increased toxic by-product N-nitrosodimethylamine (NDMA) to 33 ng/L. Dosing chloramine (common for RO biofouling control) added only up to 7 ng/L NDMA. RO tests on all pre-treated waters showed little variation to flux but oxidation significantly altered texture of RO fouling material from smooth and dense to porous and granular. Biofouling studies with model bacteria strain RO 22 (*Pseudoalteromonas* spp) showed higher organic biodegradability but biofilm analysis

revealed ozone-coagulant-CMF greatly limited extension of bacteria communities from the membrane surface suggesting oxidation reduces RO biofouling. The novel findings of reduction of RO biofouling risk with oxidation and coagulation for high flux CMF pre-treatment identified in this work need to be demonstrated on different wastewater types over longer term.

Keywords: Biofouling, ceramic membrane, coagulation, oxidation, ozone, pre-treatment, recycled water, reverse osmosis, ultraviolet /hydrogen peroxide (UVH)

## Introduction

Ceramic membranes are an alternative technology to polymeric membranes for water treatment offering superior physical integrity, chemical resistance, higher flux, and longer life [1]. However their application as a pre-treatment for reverse osmosis (RO) desalination of wastewater is unconventional. In considering ceramic membranes, high flux is important to offset their higher material cost but must be operated in a specific way to achieve this, which would impact the downstream RO plant operation. For example Dow and co-workers demonstrated that the sustainable ceramic microfiltration (CMF) membrane fluxes for treating clarified wastewater increased 2-3 fold in response to dosing with the common coagulant polyaluminium chloride (PACl) [2, 3]. Coagulation used prior to polymer membranes is already known to reduce fouling as well as to remove organic matter, particularly the large molecular weight (MW) components, being biopolymers and humic substances [4-6]. Fan et al. [7] concluded that coagulation treatment reduced organic fouling by removal of these larger-sized materials. Further, ozone used in conjunction with coagulation and ceramic membranes was observed to work together to provide >4-fold sustainable flux increases for ceramic membranes [2]. Oxidation processes such as ozonation, and ultraviolet irradiation (UV), are commonly practised as the tertiary treatments to meet appropriate water quality in reclaimed water from secondary wastewater treatment plant (WWTP) effluents for disinfection purposes, odour treatment as well as the removal of colour caused by humic substances. With their wider use in water treatment, researchers have more recently considered their specific impact on water organic fractions [6, 8] and membrane fouling [9], which is particularly useful for explaining why such high ceramic membrane fluxes can be achieved.

Studies conducted using ozone-resistant polyvinylidene fluoride (PVDF) and polysulfone (PS) membrane materials showed that using ozonation upstream of the membrane did enhance the permeate flux and reduce membrane fouling by the degradation of high molecular weight natural organic matter [10-13]. More recently, a study on polymer ultrafiltration (UF) membranes found that the mechanisms are more complex, where ozone reactions with bovine serum albumin (BSA) led to increased fouling, while reactions with alginate led to reduced fouling [9]. On top of altered organics chemistry, theories around the role of ozone regarding its ability to greatly enhance flux have focused on the role of highly reactive hydroxyl radicals ( $\text{OH}^\bullet$ ) formed by the catalytic breakdown of ozone on the ceramic membrane surface [14].

So in the case of upstream oxidation where membranes benefit in terms of performance, there is a clear alteration of the chemical properties of the water borne compounds that will impact other downstream processes. In the case of saline wastewater, low pressure membranes are widely applied prior to reverse osmosis (RO) as a pre-treatment. Normally oxidation would be applied in a water recycling scheme downstream of RO, however, it is generally understood that the mechanisms to increase hydrophilicity of organics in wastewater would be useful in controlling RO membrane fouling. Such benefits including minimising cleaning and membrane replacement, and reduced energy requirements due to reduced RO fouling, were explored in a dedicated study [15]. Membrane bioreactor (MBR) effluent was fed directly to a dual train pilot RO system with one train featuring an ozone stage, while the other fed directly by MBR permeate. The reduction to membrane fouling was demonstrated over 3000 hours of testing, showing reduced membrane permeability deterioration suggesting longer term benefits to RO membranes in terms of longevity, reduced cleaning costs and lower energy requirement [15]. Without ozone, RO flux declined by 12% while with ozone only declined by 6%. Similar beneficial effects were reported at bench scale [16]. Recent work on application of ozone and CMF followed by biologically active filtration upstream of RO for water recycling application found uniquely that RO foulants after ozone and CMF were easily removed with water rinsing [17]. This promising finding shows that in the case when ozone is applied upstream, reduced cleaning maintenance of the RO membranes is expected. The process was subsequently adopted for a 9 month potable reuse trial [18, 19]. However, these used biological processes after oxidation, may not be necessary to apply prior to RO.

Oxidation (i.e., ozone or UV) in practice is typically followed by biological filtration. Ozone breaks down larger molecular weight organic matter increasing the assimilable organic carbon proportion, favouring micro-organism growth [20]. The study by Nguyen and Roddick highlighted that the ozonation of the raw activated sludge effluent produced biodegradable dissolved organic carbon (BDOC), and biological activated carbon (BAC) filter did not completely remove those compounds [21]. Thus it is uncertain if deliberate use of BAC to prevent biofouling of downstream RO membranes would be effective. Recent work has shown that ozone and BAC application prior to ceramic membranes can have a negative impact to CMF performance compared to ozone on its own [22] suggesting that despite the BDOC removing ability of BAC, it is not useful for high CMF performance and could be avoided for pre-treatment to RO.

Disinfection by chloramines is generally practised prior to the RO process to prevent the membrane from biofouling in a conventional RO-based water recycling application [23]. Hence, despite the increase in biodegradability of organics due to ozone, the application of chloramine may assist in controlling biofouling. However, the use of chloramines can lead to the formation of disinfection by-products (DBPs), especially nitrogen-containing DBPs such as *N*-nitrosodimethylamine (NDMA) and other *N*-nitrosamine compounds [24]. On top of this, ozone is also well-known to form NDMA as a result of the oxidation of NDMA precursors [25-28]. NDMA is an important concern if the intended use of the water is limited by this compound, e.g., potable reuse. A study on ozone application upstream of RO should consider use of chloramine disinfectant and the formation of NDMA.

Therefore, it still remains unknown of the viability of using the high CMF flux arrangement (with oxidation and coagulation) as a pre-treatment to RO for saline wastewater recycling purpose, particularly in the case where no post-oxidation biological treatment stage (e.g. BAC) is used. At the same time, working towards understanding differences in RO membrane fouling (both organic and bio) of this non-traditional water recycling process compared to the more traditional approach (without oxidation prior to RO) is of more fundamental interest. Addressing these points forms the more novel feature of this work. This study therefore has the following objectives 1) to confirm reported high flux performance when ceramic membranes are coupled with coagulation, ozonation and UV/H<sub>2</sub>O<sub>2</sub> (UVH) treatment and their combinations; 2) to demonstrate the impact of the pre-treatment processes on water quality including formation of a well-known wastewater disinfection by-product, NDMA; 3) to test the influence of the pre-treatment options on RO membrane performance; and 4) to determine the potential for biofouling on the downstream RO membranes. The source water collected from a full-scale water recycling plant was used for the purpose of this work.

## Materials and Methods

### *Raw water source*

The water source used for this study was ‘Class A’ recycled water from one of Melbourne’s wastewater treatment plants (WWTP) run by the authorised operator. The WWTP receives wastewater from both domestic and industrial sources. To meet Class A specification, the incoming sewage is treated via an anaerobic and aerobic process followed by chlorination and UV treatment. The recycled Class A water has characteristics as indicated in Table 1, measured by methods described later under the ‘Water quality analyses’ section. This water is referred to as ‘direct Class A’ water hereinafter. This water is currently fed to a recently constructed salt reduction plant (SRP) which consists of a polymeric UF/RO system for water recycling application and is therefore a good model water to show an alternative ozone and CMF as a RO pre-treatment.

**Table 1:** Representative water quality indicators of Class A recycled wastewater used for this work. Method for determination described under ‘Water quality analyses’ section.

Indicator	Unit	Value
pH	-	7.7
EC	μS/cm	1700
TDS	mg/L	1240
UV <sub>254</sub>	l/m	17.9
DOC	mg-C/L	10.5
COD	mg/L	38
TN	mg/L	15.8

EC = electrical conductivity, TDS = total dissolved solids, COD = chemical oxygen demand, TN = total nitrogen

153

## 154 *Pre-treatments*

155 Pre-treatments to CMF included coagulation which was used in conjunction with ozonation or  
156 UVH. For coagulation treatment with polyaluminium chloride (PACl), 23% w/w as ( $\text{Al}_2\text{O}_3$ )  
157 from Ixom Watercare Pty Ltd, was dosed at 3 mg ( $\text{Al}^{3+}$ )/L. This dose was chosen following a  
158 series of jar tests where pin-floc was observed to start (visual observation of small flocs in  
159 solution). The required amount of PACl coagulant was added to the feed tank prior to CMF  
160 membrane, (and after oxidation by ozone or UVH when applied) without filtering the solids,  
161 to simulate the inline coagulation process used in pilot trials [2]. Ozone was generated from  
162 pure oxygen by an ozone generator (SOZ-6G, A2Z Ozone Systems Inc., USA) with an ozone  
163 production capacity of 6 g/h. Further details of the ozone dosing and analysis is provided in the  
164 Supplementary Material. During the ozone–CMF experiments, the feed water sample was  
165 ozonated for 15 minutes which was determined to be equivalent to an applied dose of 13 mg-  
166  $\text{O}_3$ /L. Residual ozone present in the ozonated samples was not quenched, and it was allowed to  
167 be in contact with the ceramic membrane surface. Ozone concentration was measured using  
168 the Indigo Colorimetric method [29]. For UVH treatment, irradiation was conducted in an  
169 annular reactor fitted with a centrally mounted UV lamp. It had a working volume of 900 mL  
170 and an average irradiated area of 464  $\text{cm}^2$ , with a path length of 1.95 cm. The UVC lamp  
171 emitted monochromatic light at 254 nm. The average fluence rate of the UVC lamp was  
172 determined to be 13.1  $\text{mW}/\text{cm}^2$  by hydrogen peroxide actinometry [30]. The effluent samples  
173 were irradiated for various times with the addition of hydrogen peroxide (1 mM). This  
174 treatment is referred to as UVH hereafter.

175

## 176 *Ceramic microfiltration (CMF)*

177 A Membralox T1-70 single channel ceramic membrane (Pall Corporation), which had a  
178 separation layer of 0.1  $\mu\text{m}$  nominal pore size, was used for CMF. The ceramic membrane had  
179 dimensions of 250 mm length and 77 mm internal diameter, a total surface area of 0.005  $\text{m}^2$ ,  
180 and was composed of a porous alumina support and selective layer made from zirconia. An  
181 example clean water flux measured for this membrane was 90  $\text{L}/(\text{m}^2\cdot\text{h})$  at 0.1 bar. Further  
182 details on the module, performance checking and cleaning are found in the Supplementary  
183 Material. The ceramic membrane was tested in dead-end, constant flux, inside-out filtration  
184 mode. Hydraulic backwashing was performed every 30 min at 3 bar via pressurised water and  
185 a series of valves. A constant flux of 130  $\text{L}/(\text{m}^2\cdot\text{h})$  was utilised for all CMF tests for the high  
186 flux operation. More details of the equipment and the method for determining constant flux are  
187 described in the Supplementary Material.

188

189 Filtration performance was evaluated by using indicators such as fouling rate and backwash  
190 effectiveness (fouling reversibility). The rate of foulant accumulation on the membrane or  
191 fouling rate over time was described as the change in transmembrane pressure (TMP) per unit

time ( $dTMP/dt$ ) on a per cycle basis. The change in fouling rate (%) was calculated by the following equation [31]:

$$\text{Change in fouling rate (\%)} = \frac{\left(\frac{dTMP}{dt_n}\right) - \left(\frac{dTMP}{dt_1}\right)}{\left(\frac{dTMP}{dt_1}\right)} 100 \quad (1)$$

where the fouling rate of the final filtration cycle (cycle  $n$ ) was compared to the initial filtration cycle (cycle 1).

To assess membrane performance data between filtration and backwash cycles, the following method was used based on the unified membrane fouling index (UMFI) developed by Huang et al. (2008) [32] and Nguyen et al. (2011) [33]. All TMP data points were used to calculate specific flux or permeability,  $J_s$ , ( $L/(m^2 \cdot h \cdot bar)$ ) as follows:

$$J_s = J/TMP = 1/\mu (K_{mem} + k_{total} V) \quad (2)$$

Where  $\mu$  is viscosity,  $K_{mem}$  is the resistance of the clean membrane,  $k_{total}$  is the total resistance (membrane and fouling resistances), and  $V$  is the specific volume ( $L/m^2$ ).

For a clean membrane, at  $V = 0$ ,  $(J/TMP)_0 = 1/\mu K_{mem}$ . Membrane performance can be represented in normalized form,  $J'_s$ , by dividing  $J/TMP$  at any specific volume by the initial (or clean membrane) condition according to the following equation:

$$J'_s = \frac{\left(\frac{J}{TMP}\right)_V}{\left(\frac{J}{TMP}\right)_0} = \frac{1}{1 + \frac{k_{total} V}{K_{mem}}} \quad (3)$$

or

$$\frac{1}{J'_s} = 1 + \left(\frac{k_{total}}{K_{mem}}\right) V \quad (4)$$

Different fouling indices could be calculated from plotting a graph of  $1/J'_s$  versus  $V$ . Hydraulic irreversible fouling index (HIFI) can be calculated using the starting TMP data point after each backwash cycle. HIFI is related to the fouling resistance and a low HIFI represents a low rate of membrane fouling while a high HIFI indicates greater membrane fouling rates.

#### *RO feed chlorination, membrane fouling loading tests and SEM analysis*

A dose of preformed chloramine was added to each filtrate of the CMF as per the conditions established on current RO systems (approximately 4 mg/L) and similar to previous studies [16, 34]. For antiscalant dosing, the commonly used Flocon 260 was dosed at 3 mg/L to the solution to represent realistic application to RO feeds.

The RO membrane fouling loading test for the pre-treated waters was performed on a DOW FILMTEC BW-30 membrane. The schematic diagram of RO membrane filtration set up and

further details of the operation are found in the Supplementary Material. The effective membrane area was 0.014 m<sup>2</sup> and cross flow velocity of 0.2-0.3 m s<sup>-1</sup>, and run in batch concentration to achieve a final volume recovery of 80%. Pressure of the feed was set to 10 bar.

SEM was employed to investigate the morphology of the membrane surface and accumulated fouling material. The SEM images were produced using a NeoScope JCM5000 (JEOL, Japan) with a 10 kV electron beam. To improve the imaging of the samples, the membranes were gold coated using a Neo coater MP-19020NCTR (JEOL, Japan) prior to the observations.

### *Water quality analyses*

The feed samples before and after pre-treatments were analysed for pH, electrical conductivity (EC), ultraviolet absorbance at 254 nm (UV<sub>254</sub>) and dissolved organic carbon (DOC). Some indicators were measured and shown only in Table 1 to give an overview of the water quality (i.e. includes also TDS, COD and TN). pH and EC were determined using a HACH Sension 156 handheld meter. TDS was determined using Standard Method 2504. UV<sub>254</sub> was measured using an HACH DR 5000 spectrophotometer. Specific UV absorbance (SUVA) was determined by dividing UV<sub>254</sub> by the DOC concentration. DOC and TN concentrations were measured using a total organic carbon analyser (TOC-V<sub>CPH</sub>/CPN, Shimadzu, Japan). DOC fractionation was performed by liquid size exclusion chromatography with organic carbon detection (LC-OCD) using a LC-OCD model 8 system (DOC-Labor Dr. Huber, Karlsruhe, Germany) at Technische Universität Dresden, Germany. LC-OCD enables the characterisation and quantification of DOC fractions. Details of the method have been published elsewhere [35, 36] and a further summary can be found in the Supplementary Material.

The concentration of biodegradable dissolved organic carbon (BDOC) of the waters before and after treatments was determined using the method of Joret and Levi (1986) [37] and modified according to the method reported by Volk et al., (1993) [38]. Briefly, a sample of 300 mL was exposed to washed biologically active sand (100 ± 10 g) for 7 days under aerobic conditions (3 litres of humidified air per hour). The BDOC was calculated as the initial DOC minus the lowest DOC recorded over the 7-day incubation period.

The chloraminated (4 mg/L as Cl<sub>2</sub>) ceramic membrane permeate samples were analysed for N-nitrosamines analysis after 4 mg/L chloramine dosing. N-nitrosamines were analysed by solid-phase extraction (SPE) followed by gas chromatograph with mass spectrometer detector (GC-MS), based on the method of Charrois et al. (2004) [39] with minor modifications. Further details of the method are found in the Supplementary Material section.

### *Bioassay and accelerated RO biofouling tests*

Biofilm assays were conducted using a crystal violet assay method [40]. A model biofouling bacterial strain, RO 22, was used to evaluate the biofouling potential of the treated waters. RO 22 is a strain of *Pseudoalteromonas* spp isolated from a full scale SWRO plant [41]. Additional details of the organism and methods are found in the Supplementary Material. A

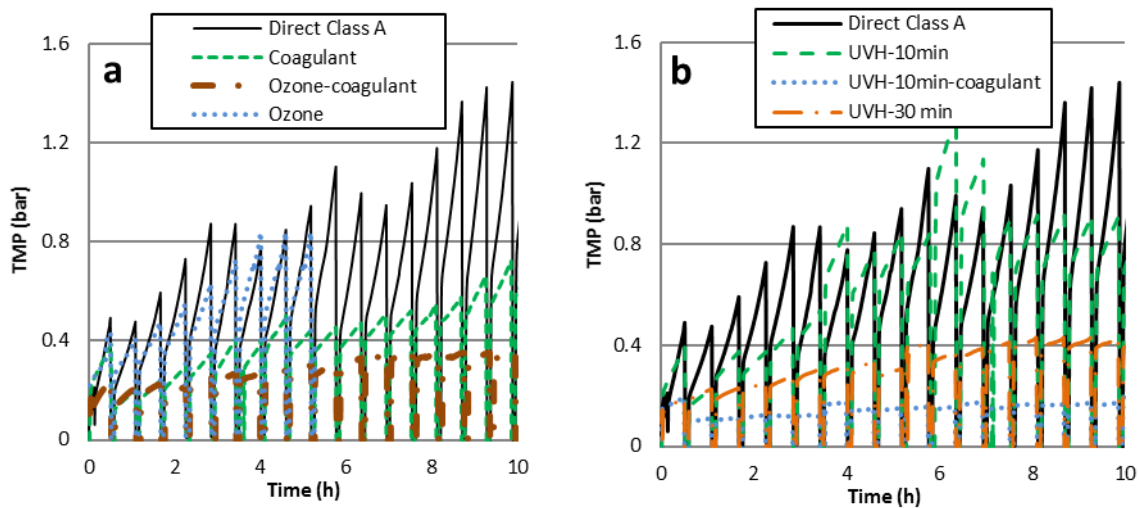


single colony of isolate RO 22 was inoculated into 10 mL of tryptone soy broth (TSB) and mixed well. 200  $\mu$ L of culture broth was pipetted into each well in a 96-well microtitre plate, sealed and incubated for 48 hours at room temperature. Optical density of the wells to determine cell density, then removal of solution and addition of crystal violet to measure biofilm thickness, were both measured using light absorbance. For the accelerated biofouling tests, RO feeds were spiked with RO 22. The biofouling tests were conducted for a total of 21 h. After 21 h of RO filtration tests, a dose of preformed chloramine (approximately 4 mg/L) was added to the feed reservoir to observe effects due to the chloramine addition. At the end of each biofouling experiment, the membrane coupon was carefully removed and the membranes were preserved for confocal microscopic observation.

## Results and Discussion

### *Ceramic membrane operation performance*

Figure 1 shows the transmembrane pressure (TMP) profile of each process operated at constant CMF flux of 130 L/(m<sup>2</sup>.h). Figure 1a shows TMP profiles during filtration of direct Class A water, and the same waters after adding coagulant, ozone and their combination. The results showed that feeding the Class A water directly to the ceramic membrane led to rapid fouling as observed from the rapid rise in TMP to 1.4 bar within 10 h of filtration time for 130 L/(m<sup>2</sup>.h) flux operation. The fouling rate for direct Class A water increased from 0.64 to 1.6 bar/h at the first (1<sup>st</sup>) and last (17<sup>th</sup>) filtration cycle respectively.



**Figure 1:** TMP rises as a function of time for direct Class A feed, coagulant feed, ozone feed and ozone-coagulant feed (a) and UVH-10 min, UVH-30 min and UVH-10 min-coagulant feed (b). All fluxes were fixed at 130 L/(m<sup>2</sup>.h).

Under the same operating conditions, when 3 mg (as Al<sup>3+</sup>)/L of polyaluminium chloride (PACl) coagulant was dosed prior to the membrane, TMP rise was reduced. Compared to

direct Class A feed, TMP rose more slowly and approached 0.68 bar for the same volume of water filtered. The reduction was mostly associated with reduced rises between backwashes. When ozone only was added to the feed water, the TMP showed a reduced rise between backwashes initially. Using either coagulant or ozone, the gradual build-up of TMP over the course of the run appeared similar to direct Class A water feed. In the case of ozone, it has been recently reported that reduced TMP rise between backwashes could be due to the reduced flow resistance in the oxidised organic matter accumulated on the membrane surface [9]. However, when ozone and coagulation are combined, both overall and between backwash TMP rises were greatly reduced. The finding is supported in pilot trials of CMF on recycled wastewater where ozone was observed to reduce the TMP rise during filtration while coagulation reduced TMP rise after each backwash [1, 2]. Using the data in Figure 1 and Equation 1, change in fouling rate can be calculated to compare performance. The fouling rate increases by up to 150% during the 10 h filtration period for the direct Class A water. In the case of ozone, this reduced to 112%, indicating for the chosen flux of 130 L/(m<sup>2</sup>.h) fouling was increasing. When coagulant was instead applied, the change in fouling rate was similar. However the change in fouling rate reduced significantly to 27% with the combined ozone-coagulant feed to the ceramic membrane. Coagulant and ozone thus inhibit the need for chemically enhanced backwashes which remove irreversible foulants that cause accelerated fouling rates [3]. The result here confirms the well-known effect of greatly enhanced sustainable fluxes following coagulation and ozonation and the filtrate is suitable for downstream RO processing.

Figure 1b demonstrates the TMP profile of each process operated at 130 L/(m<sup>2</sup>.h) flux for direct Class A feed and feed pre-treated with UVH and combined UVH and coagulation process. Longer UV treatment time from 10 minutes to 30 minutes greatly reduced TMP rise between backwashes, which could be due to similar reasons of reduced filter cake resistance as observed for ozone [9]. Spikes in TMP were observed for some filtration cycles from the 10 minute UVH (UVH-10 min, Fig. 1b), exceeding the TMP of direct Class A feed between 3 and 7 hours. This however was considered to be due to experimental issues, for example air accumulation in the membrane tube which was removed during backwash. Importantly however, TMP rise rate between backwashes was consistently lower than direct Class A highlighting the reduced filter cake resistance. Coagulation was only added to the 10 minute UVH case as it showed a near complete removal of TMP build-up in the 10 hour test period. The effect appears similar to that for ozonation – coagulation treatment. The change in fouling rate was approximately 20%, which is similar to the ozone-coagulant treated water.

The HIFIs shown in Table 2 show the normalised quantitative differences between the CMF filtration scenarios, where any oxidation process leads to significantly reduced fouling when used in conjunction with coagulation. Although either may be suitable, previous studies directed to RO membrane fouling benefits found UVH more expensive than ozone [15] suggesting the importance in considering cost in deciding to use either ozone or UVH.

**Table 2.** HIFI on CMF for each pre-treated water

<u>Pre-treatment</u>	<u>CMF HIFI (m<sup>2</sup>/L)</u>
Direct Class A	2.75
Coagulation	2.08
Ozone + Coagulation	1.25
UVH-10min + Coagulation	0.37

### *Ceramic membrane treatment performance*

The measured quality indicators of the untreated Class A water feed and the various pre-treatments options are shown in Table 3. Originally, the feed water showed relatively high DOC and low UV<sub>254</sub> absorbance. DOC removal was <10% for coagulation, ozone or UVH, but when coagulation was combined with oxidation, DOC removal increased to 10% for UVH+coagulation, and 18% for ozone+coagulation possibly by ozone enhanced coagulation effects [42]. Coagulation reduced UV<sub>254</sub> absorbance by only 13%, but any combination with oxidation led to significant reductions between 44% and 63%, where the highest was measured in the ozone cases.

**Table 3.** Measured water quality indicators after various pre-treatment options prior to CMF

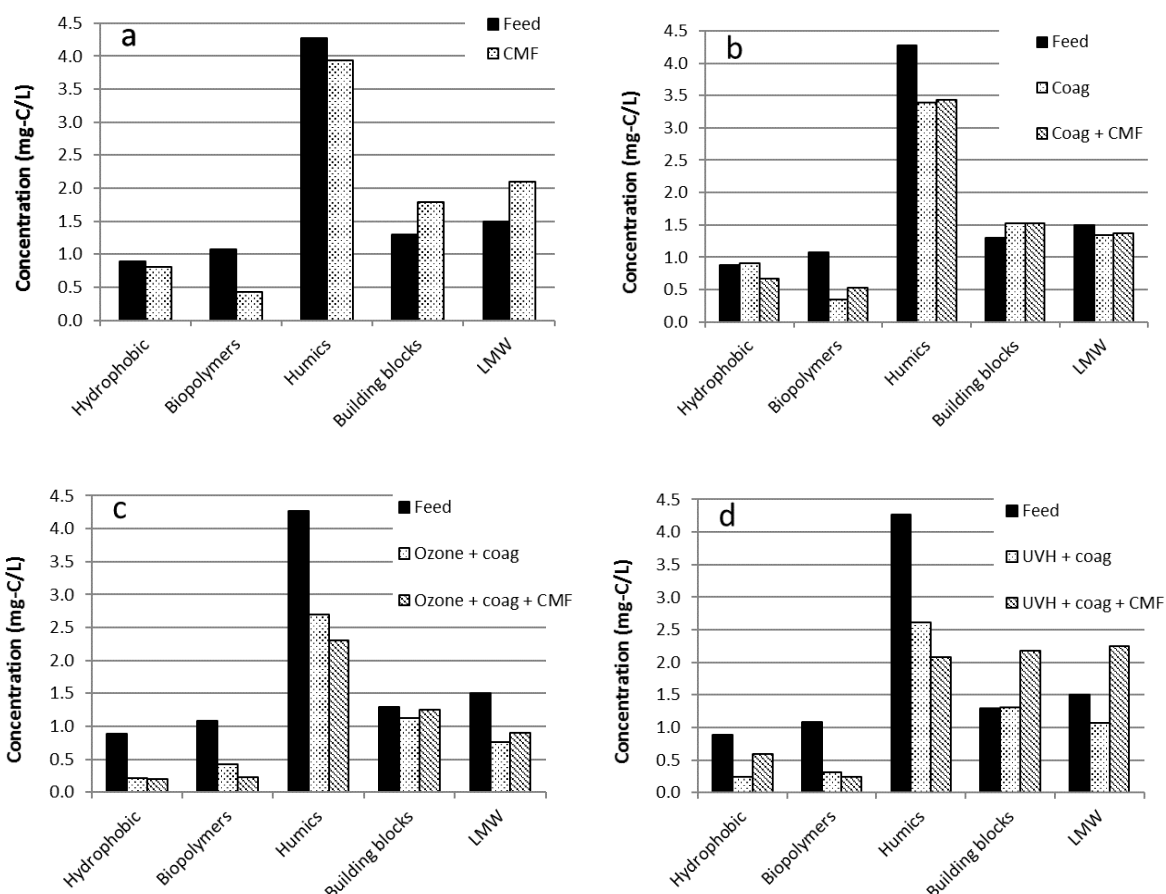
<b>Pre-treatment</b>	<b>UV<sub>254</sub> (1/m)</b>	<b>pH</b>	<b>DOC (mg/L)</b>	<b>SUVA (L/(mg·m))</b>
None (direct Class A)	16	7.43	9.1±0.4	1.74
Coagulation	14	7.53	8.3±0.3	1.70
Ozone	7	7.44	8.4±0.6	0.88
Ozone+coagulation	6	7.39	7.5±0.1	0.78
UVH	9	7.51	8.6±0.2	1.02
UVH+coagulation	8	7.57	8.2±0.4	1.02

Organic fractions within the various pre-treatment options stages were analysed more closely by LC-OCD and the results are shown in Figure 2. The LC-OCD analysis enables the quantification of organic matter fractions including biopolymers (MW>>20,000 g/mol), humic substances (MW~1000 g/mol), building blocks (MW 300-500 g/mol), low molecular weight (LMW) substances (MW <350 g/mol) which are the sum of low molecular weight neutrals and low molecular weight acids. The results show that feed water dissolved organic material consisted of 47% humic-like substances, 17% LMW substances, 14% building blocks, 12% biopolymers and 10% hydrophobic organics. Membrane filtration (Figure 2a) removed mostly the biopolymer proportion (60% removal) due to their high molecular weight and their sticky

properties. Humic substances were almost similar in concentration in the membrane permeate and the feed. They can readily pass through the membrane pores of 0.1  $\mu\text{m}$  in size. A small fraction of large humic substances might be retained due to tortuosity effects. For the smaller components (building blocks and LMW organics) slight increases were observed, which cannot be explained and may be due to sample handling.

Considering the results with the pre-treatment options prior to CMF biopolymer removals of 68%, 60% and 71% for coagulation, ozone+coagulation and UVH+coagulation treatments were observed respectively. Biopolymers are readily removed by coagulation [6] whereas oxidation processes result in a breakdown of the large molecules and production of smaller molecules which are harder to be removed by coagulation observed only in the case of ozone. At the first glance it seems that ozonation was more effective in breaking down the biopolymers. However, due to the single sample analysis, it is questionable whether that effect is significant. Application of CMF to complete these pre-treatment options did not contribute to large additional differences in the biopolymer concentrations.

Removal of humic substances prior to CMF also occurred in all cases, but was highest in both oxidation cases (Figures 2c and d) at about 40% as opposed to 21% for just coagulation. Application of CMF did not appear to offer humic substances removal applied directly to the feed or after coagulation, but some additional removal occurred after oxidation, leading to a total humic substances removal of about 50% for both ozone and UVH cases. In the case of building blocks, no noticeable concentration changes due to the pre-treatment prior to CMF were observed. However, unexpectedly concentrations of the LMW fractions were lower for ozone and UVH than coagulation alone. Previous studies on ozone and UVH reaction with wastewater organics coming from the conventional activated sludge process had shown increases in the proportion of LMW acids [8]. The LMW acids make up the LMW fractions presented in Figure 2, and no increase was observed in our case potentially due to the lower relative doses of ozone and UVH where LMW acid formation is lower. Also, while the concentration of humic substances was similar in their work, our water contained less of the other organic fractions and therefore had a different initial organic profile highlighting the differences between various wastewater sources.



**Figure 2:** DOC fractionation after various stages within the pre-treatment options of CMF only (a), coagulation+ CMF (b), ozone+ coagulation + CMF (c) and UVH+ coagulation + CMF (d).

When pre-treatments were used with CMF the membrane generally showed little change to the relative profile of organics. This is expected due to the relatively large pore size (0.1  $\mu\text{m}$ ), but in the case of CMF on its own (Figure 2a), the larger biopolymer molecules were rejected by the membrane. As mentioned earlier, oxidation assisted the CMF to remove additional humic substances. The only other exception was in the case of UVH, which showed unusually high levels of building blocks and LMW organics in the permeate compared to the CMF feed. The reason for this is unknown since the membrane is not expected to increase any organic fraction unless it could come from particle organic matter as a result of advanced oxidation UVH. However this would be the case if also seen in the UVH+coagulation sample. Contributions to LMW fraction by oxidation of other dissolved organic fractions is also known [8], but is not expected to have occurred in just this test with CMF, particularly since this effect occurs only in very high UVH doses as compared to here. The potential for adsorbed organics to be released from the membrane due to oxidation is also ruled out because the samples were taken in batch from the oxidation process to the CMF test unit (enough time for residual oxidants to be consumed prior to contacting membrane). This increase is therefore unexpected and because only one sample was analysed, it may relate just to the experimental or preparation of this sample.

417

418 Considering differences between ozone and UVH, González et al [8] reported ozone and UVH  
419 techniques lead to different impacts on the organic fractions as also assessed by LC-OCD. In  
420 the case of ozone in Figure 2c, the effect in addition to coagulation (Figure 2b) showed its  
421 selective nature participating in removal of humic substances and LMW organics. Low  
422 biopolymers in CMF and coagulation+CMF cases were consistently lower in oxidised samples  
423 hence the oxidation process (including peptide bond cleavage and depolymerisation of  
424 polysaccharides) did not lead to any observed breakthrough of biopolymer substances (proteins  
425 and polysaccharides) to the CMF permeate. Ozone is also known to react preferably with the  
426 highly aromatic humic substances, and the slightly aromatic and hydrophobic LMW neutrals  
427 [43]. As shown in Figures 2c and d, the oxidation processes can also be seen to increase the  
428 hydrophilicity of the DOC, where no large removal of the hydrophobic organic fraction was  
429 observed in CMF and coagulation+CMF cases, while oxidation led to 34% to 77% removal  
430 across all the pre-treatment steps. In terms of aromaticity, oxidation led to a large reduction as  
431 indicated by the SUVA results shown in Table 3. Although the humic substances were the  
432 mostly dominant organic fraction after pre-treatment, the oxidation processes are expected to  
433 deplete the aromaticity of these organics by attack of double bonds and aromatic rings but not  
434 to cleave them leading to structure loss, unless high oxidation doses are used where LMW  
435 fractions are observed to increase [8]. The resistance of humic substances to oxidation has been  
436 proposed to be associated with steric obstruction preventing cleavage of the core molecular  
437 bonds of humic structures [44]. These changes in chemical composition, together with their  
438 lower overall relative abundance, could lead to the reduced fouling of RO membranes observed  
439 previously [15, 16] and is expected to greatly alter the biofouling propensity. Both will be  
440 explored later in this paper.

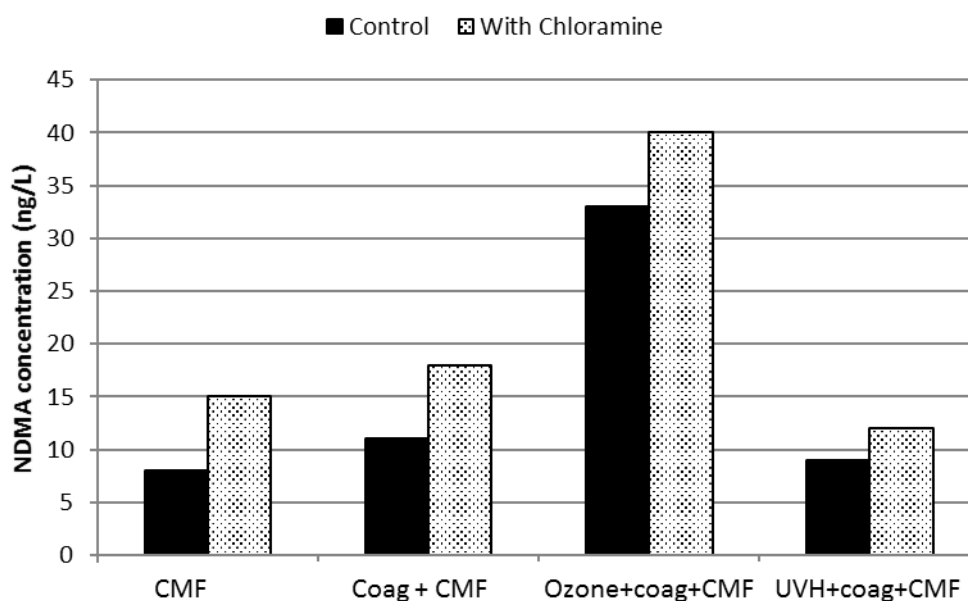
441

#### 442 *N-nitrosamines analysis of ceramic membrane permeate*

443 Figure 3 shows NDMA concentrations observed after the various ceramic membrane pre-  
444 treatment processes. NDMA was detected in the CMF only treated water at 8 ng/L.  
445 Coagulation+CMF showed a slightly higher NDMA concentration at 11 ng/L, while  
446 ozone+coagulation+CMF caused a large increase to 33 ng/L. UVH+coagulation+CMF on the  
447 other hand showed a concentration of 9 ng/L, being similar to the CMF or coagulation+CMF  
448 treated waters. Chloramine dosing tended to increase NDMA concentrations in any sample  
449 by 7 ng/L, except for UVH which increased only by 3 ng/L. Ozone is important to achieving  
450 the desirable high CMF fluxes, but clearly its impact to form NDMA is more critical than  
451 chloramine which may be used to control RO membrane biofouling. Oxidation is known to  
452 greatly reduce the potential for NDMA formation in drinking water application [45] however  
453 ozone also induces NDMA formation when applied to wastewater. 7 out of 8 wastewater  
454 treatment plants surveyed for DBP formation associated an increase in NDMA as a result of  
455 the ozonation stage [46]. Biological filtration following ozone assisted in removing formed  
456 NDMA. In the same work, the O<sub>3</sub>/DOC mass ratio was analysed, where plants showed ratios  
457 ranging from 0.2 to 1.5 w/w. In our case, the ratio was 1.4 w/w as ozone dose was 13 mg-

O<sub>3</sub>/L and DOC was 9.0 mg-C/L, which is at the higher end of their reported range. While this higher DOC supports the ability for ozone to form NDMA, their survey of various plants found no conclusive link to O<sub>3</sub>/DOC mass ratio, where instead NDMA formation is more likely dependant on the presence of precursors in the wastewater and the extent of treatment by the upstream treatment plant. Krasner et al. [47] reported that NDMA can form during ozonation of a limited group of tertiary amine precursors present in drinking water (including wastewater impacted source waters), although the association between ozonation and NDMA formation has not yet been found.

The issue of increased NDMA formation from application of ozone upstream of RO has been considered previously [15], where the reduced NDMA formation potential by ozone was not offset by its role in producing NDMA. It was concluded that this will be important in applications where NDMA concentration is monitored, for example potable reuse. Use of UVH to avoid NDMA in such applications may offset the potentially higher costs mentioned earlier. Optimisation of the UV and H<sub>2</sub>O<sub>2</sub> dose may find a means to achieve the desired oxidation with lower energy [34]. The result in Figure 3 suggests that for consideration of the use of the recycled water, the choice of oxidation to achieve high CMF fluxes will have a strong impact on NDMA formation while chloramine dosing will have less of an effect.

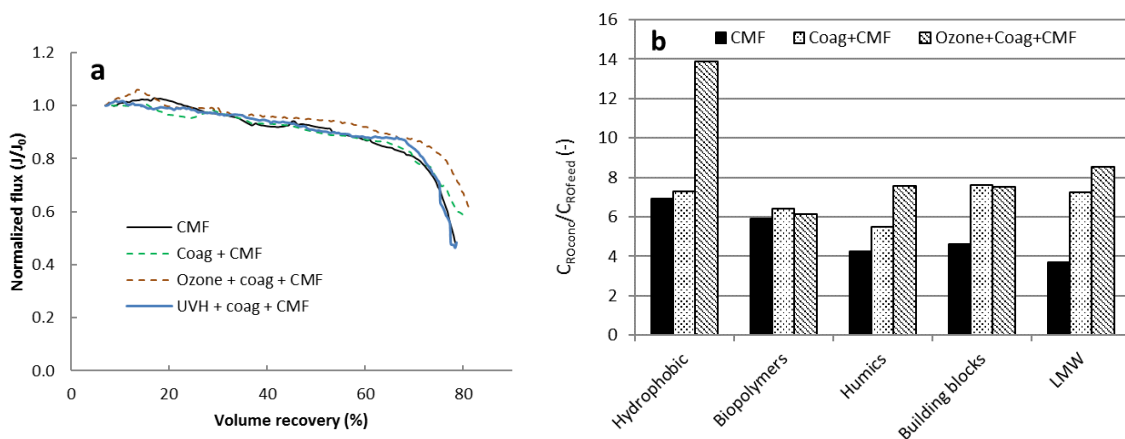


**Figure 3:** NDMA concentration measured for both control and chloraminated samples of feed waters (dosed with 4 mg/L pre-formed chloramine) pre-treated with CMF, coagulant+CMF, ozone+coagulant+CMF and UVH+coagulant+ CMF. Chloraminated waters used as RO feeds.

*Impact of CMF pre-treatments on downstream RO*

## RO performance during fouling loading

The effect of fouling of the RO membranes pre-treated by CMF, coagulation+CMF, ozone+coagulation+CMF and UVH+coagulation+CMF was compared and the data is shown in Figure 4. In these cross-flow batch concentration runs, similar flux decline results were observed for all pre-treatment modes indicating the accumulated fouling under any water feed did not show differences in resistance of water flux through the membrane. The decline experienced by all samples is likely due to the increasing salinity and in turn osmotic pressure which reduces the flux. A slight benefit to performance, however, was observed for the ozone+coagulation+CMF pre-treated water where the drop off in flux occurred at a slightly higher recovery than the other samples. Previous research showed strong benefits of ozone or UVH to prevent increasing flux resistance through the RO membrane between 70 and 120 hours of testing [34]. A key difference in our study was application of oxidation prior to membrane filtration, where the CMF must follow from oxidation in order to achieve the high flux effect. Oxidation in our case can therefore react with additional organics (i.e. biopolymers) that are removed by membrane filtration (Figure 2), and may have minimised the differences to RO fouling resistance. However, unlike the previously reported benefits of oxidation and the slight benefit from our testing with ozone, we did not see any benefit to RO membrane flux as a result of UVH. Therefore based on our short term test result, applying oxidation prior to CMF shows no significant advantage to RO membrane flux as a result of altered organic/inorganic fouling properties. .



**Figure 4:** Normalized flux decline versus permeate recovery (%) for various feeds (a) and ratio of concentrations of the various organic groups between RO concentrate and RO feed,  $C_{ROconc}/C_{ROfeed}$ , with various pre-treatments determined by LC-OCD (b).

The RO test aimed to simulate a RO plant where in a single housing, a series of elements (e.g., 7 elements) operating at effectively the same pressure at a given moment in time within the vessel have decreasing fluxes along the length of the vessel as a function of the concentration (water recovery). Lead elements have higher flux than tail elements. However, the fouling on the membranes in the bench setup differs from the real plant in that it is the same membrane



tested from the initial water rejected (lead element) to the final rejection (tail element). Fouling, (including biofouling) on lead and tail elements has been investigated in pilot sea water and wastewater RO plants showing very different behaviour unique to the fluxes and brine concentrations that differ greatly along the membrane pressure vessel [48]. Respecting these differences that are more difficult to replicate at bench scale, the test conducted here conveniently and quickly shows the average fouling across all elements. Tail elements with higher brine concentrations (70% recovery) were shown to have higher degree of mineral scaling over biofouling [48]. Therefore, the foulant here represents accumulated organic and inorganic substances across the entire rejection range.

EC for both RO concentrate and permeate at 80% volume recovery are summarised for each pre-treated water type (with CMF) in Table 4. Prior to testing with samples, benchmark rejection of 99.5% was confirmed using 2000 ppm NaCl at 15.5 bar for 15% recovery. Looking at the permeates from sample testing, higher EC from oxidised samples was observed which was related to the higher concentrations of the feeds used and not due to changes as a result of ozone. This finding is supported by previous pilot trials which found no change in salt transport through the membrane due to altered chemical properties as a result of upstream ozone treatment [15]. They also concluded that the ozone did not deteriorate the RO membrane as a result of this observation after 3000 hours of pilot testing due to rapid quenching of ozone by the organic and mineral components of the wastewater such that no harmful residual entered the RO membrane unit.

**Table 4:** EC for RO feeds from various pre-treatments, as well as the concentrate and permeate collected at the end of the runs

Pre-treatment	RO feed ( $\mu\text{S/cm}$ )	RO concentrate ( $\mu\text{S/cm}$ )	RO permeate ( $\mu\text{S/cm}$ )
CMF	1298	5470	82
Coagulation+CMF	1298	6660	76
Ozone+coagulation+CMF	1326	6130	158
UVH+coagulation+CMF	1348	5560	102

An increase in permeate conductivity due to charged organics from oxidation diffusing through the membrane was also not likely, where previous testing on two wastewater sources found similar or slightly lower DOC concentrations in RO permeates for ozone+CMF treated wastewaters compared to untreated wastewater feeds [17]. Due to the focus in this work on the process train performance and membrane fouling, further analysis of the RO permeate was not conducted but would be an interesting suitable topic for future work to compare the differences between the processes.

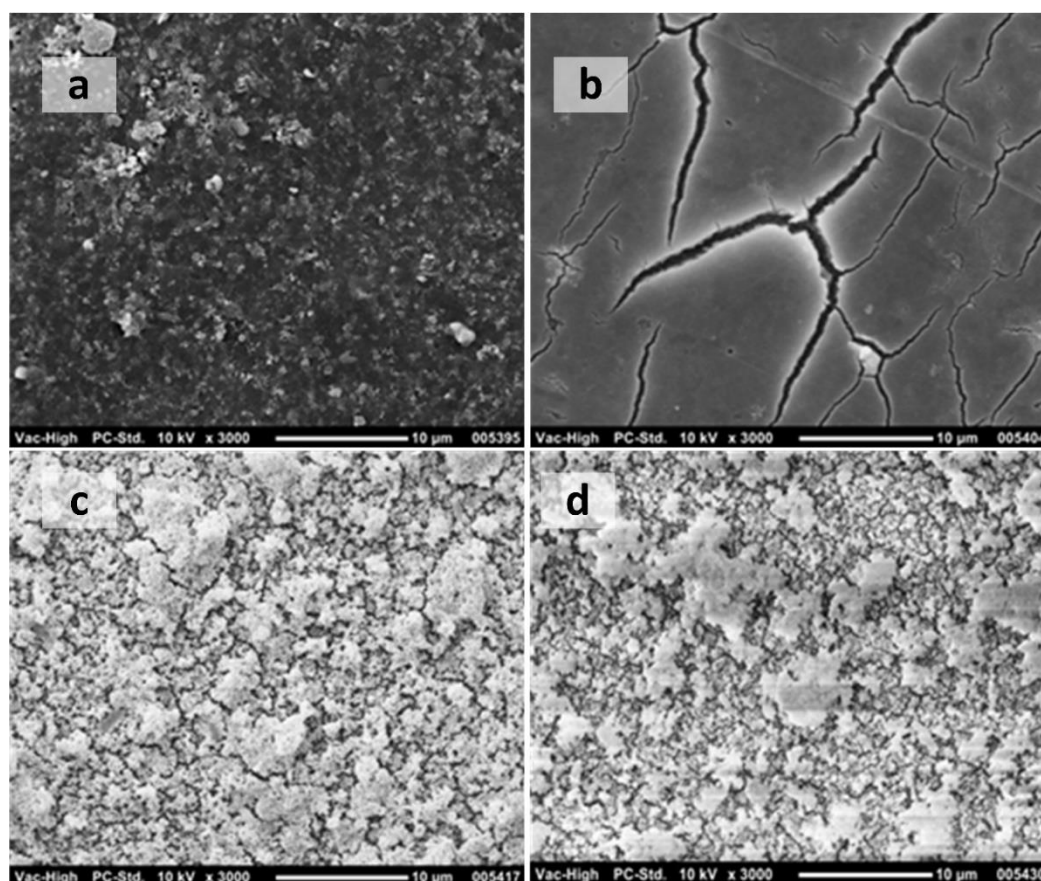
Potential depositions on the RO membrane could be explored by observing the organic fractions concentration factors presented in Figure 4b (concentration ratio in the RO

concentrate,  $C_{ROconc}$ , to the feed,  $C_{ROfeed}$ ). Factors of 5 would be expected for volume recovery of 80% if little permeated the membrane, or came out of solution (either by precipitation in the system or deposition on the membrane). However, the ratios were instead mostly around 7 and lower, suggesting that the recovery based on the concentration ratio was closer to 85%. The exception to this was the very large ratio (13.9) for the hydrophobic organics in ozone+coagulation+CMF pre-treated water which will be discussed later.

The hydrophobic fraction ratios was similar for CMF only (6.9) to coagulant + CMF (7.3), and close to the concentration ratio around 7 indicating little permeation or deposition of these fractions. Ozone+coagulation+CMF showed a very high ratio (13.9) where  $C_{ROfeed}$  was low due to oxidation, but increased more than the RO concentration factor. It cannot be concluded from the present data if this was due to an effect to increase hydrophobic property of organics due to concentrating, or sensitivity of the ratio to low feed concentration of hydrophobic organics (0.2 mg-C/L). Looking at biopolymers, all ratios were much closer between 5.9 and 6.4, and lower than the concentration ratio of ~7. Due to their high molecular weight, biopolymers are not expected to permeate the membrane and therefore it is suggested they are deposited within the concentrate cycle including depositing on the membrane. In all pre-treatment cases, it is possible that biopolymers contributed to membrane organic fouling. Humic substance concentration ratios on the other hand were different in all RO pre-treatment cases. For CMF only, a lower ratio of 4.3 for humics can be seen suggesting their limited ability to concentrate and potential to deposit on, or diffuse through, the membrane. With coagulation+CMF, then ozone+coagulation+CMF, progressively higher increases in humic substance proportions reaching the concentration expected for reduced deposition, or complete rejection, by the membrane. This is especially the case when they were reacted by ozone where it approached the system concentration ratio of ~7. Building blocks and LMW organics, which showed higher deposition on, or passage through, the RO membrane with just CMF and in turn lower increase in the RO concentrate, suddenly reached the full concentration factor apparently due just to coagulation. Their association with coagulant appears to have inhibited their ability to attach to or transport through the membrane, and they instead concentrate in the RO brine.

SEM images of the membrane surface taken at the end of the RO treatment process are shown in Figure 5. An SEM image of the original RO membrane is presented in the Supplementary Material Figure S3 for reference. The images of the fouled RO membranes show that each treatment type used in conjunction with the ceramic membrane led to very different structural features of the fouling layer deposited on the RO membranes. CMF pre-treated water led to a uniformly grainy textured material, with particles around 0.55  $\mu\text{m}$  and less in diameter appearing embedded within a smooth polymeric-like material. The particles may have originally been small enough to permeate the 0.1  $\mu\text{m}$  CMF membrane, and agglomerate on the RO membrane surface. When coagulation+CMF pre-treated water was used, a very smooth texture appeared with no visible particles. It appears the application of coagulant facilitated removal of the particulate matter by the ceramic membrane. The fouling layers of the ozone+coagulation+CMF and UVH+coagulation+CMF pre-treated water were similar to each other and very different to CMF or coagulation only pre-treatments. They uniquely showed a

grainy texture of highly variable agglomerates of  $<1\ \mu\text{m}$  to several  $\mu\text{m}$  in size. It appears that the material that permeated through the CMF after oxidation by ozone or UVH formed aggregated structures rather than a smooth, continuous gel layer as observed for coagulation or direct filtration of the wastewater. A different result was observed on bench and pilot tests where RO membranes were fouled with ozonated MBR filtrate. While dense fouling layers were also observed with original (not oxidised) wastewaters, the fouling layer from the ozonated wastewater was also more open [16], but did not show a grainy texture as observed here. This may relate to differences in the wastewater, ozone dose approach, and final water recoveries. Aggregation of biopolymers following ozonation has been observed by Yu et al for synthetic water systems [9]. The aggregates were used to explain mechanisms of reduced or increased fouling of a 10 to 20 nm UF membrane as a result of increasing sizes of alginate and model protein BSA, respectively. While the focus of their work was the fouling of the UF membrane, their results showed the aggregation mechanisms which could relate to organics in the CMF permeate which will be fed to RO.



**Figure 5:** SEM images of fouled RO membranes, including membrane fouled with CMF only treated water (a), with coagulation + CMF (b), ozone + coagulant + CMF and (c) UVH + coagulation + CMF (d). Original membrane without fouling shown in Supplementary Material Figure S3.

At the end of the RO experiments (i.e., after 80% volume recovery), the membranes were rinsed with clean water and clean water flux was measured. When rinsing with clean water the loosely attached foulants could be expected to detach during hydraulic cleaning, and reversible fouling is a measure of flux that could be restored after hydraulic cleaning. Clean water fluxes of 8.6, 20, 16 and 13 L/(m<sup>2</sup>.h) were measured for membrane previously exposed to CMF, coagulation+CMF, ozone+coagulation+CMF and UVH+coagulation+CMF pre-treated feeds, respectively. The higher clean water flux represents higher fouling reversibility, comparing to the new membrane clean water flux of 22 L/(m<sup>2</sup>.h) as shown in Figure S4 in the Supplementary Material. Therefore, the action of coagulation (together with ozonation and UVH treatment), on the organics leads to greater fouling reversibility on the RO membranes. Coagulation on its own was most superior in reversing fouling, while either oxidation in addition to coagulation showed less reversibility. This may be due to texture differences of the oxidised foulants as shown in Figure 5, or potentially their chemical differences, where less fouling material was removed due to simple water rinsing. The ability of clean water to more easily remove fouling matter after ozone application (without coagulant) was also found by Zhang et al. [17]. In practice, a treatment process that leads to high membrane fouling reversibility suggests that less intensive chemical cleaning will be required for the downstream RO membranes.

#### Biofouling potential tests

Table 5 shows the BDOC test results specifically for waters pre-treated to be fed to RO. No significant change in BDOC was observed between CMF or coagulation+CMF pre-treatment. BDOC however increased after ozone, ozone+coagulation, UVH and UVH+coagulation prior to CMF pre-treatments. This indicates that the oxidation processes used to increase CMF flux lead to increased biodegradability, and in turn increases the potential for biofouling if fed directly to RO as proposed in our work. It is more common in practice for biological filtration to follow oxidation especially to avoid the biofouling risk [49]. For example a biologically activated carbon (BAC) filter was installed between the ozone-ceramic membrane and RO processes, and found to remove 30% to 50% of the DOC [19] and no biofouling issues were identified over 9 months of operation [18]. However, the focus of this work was to assess the potential for biofouling where BAC pre-treatment before RO may not be required, and increased ability to assimilate organics may not directly correlate to the ability for a biofilm to form on the RO membrane.

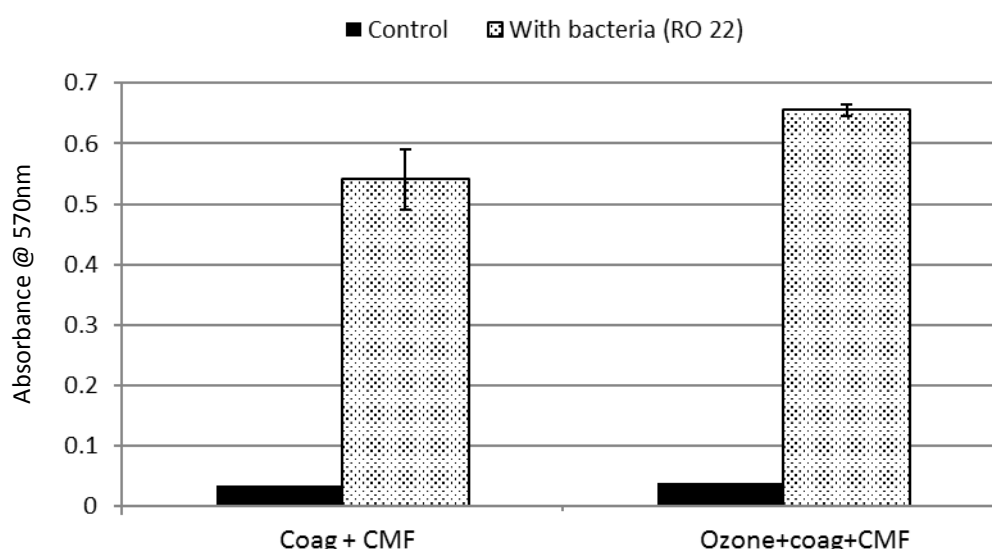
**Table 5.** Biodegradability of water samples after various pre-treatments, including those used as feed to RO

Pre-treatment	BDOC mg/L	RDOC* mg/L	DOC mg/L	BDOC % of DOC
CMF	0.50	7.83	8.33	6.0
Coagulation+CMF	0.49	7.87	8.36	5.9
Ozone +CMF	1.34	6.46	7.80	17.2
Ozone+ coagulation +CMF	1.17	6.11	7.28	16.1
UVH+CMF	1.11	6.82	7.93	14.0
UVH+coagulation+CMF	1.08	6.75	7.83	13.8

\*RDOC = Refractory dissolved organic carbon = Total DOC – BDOC

### Bioassay results

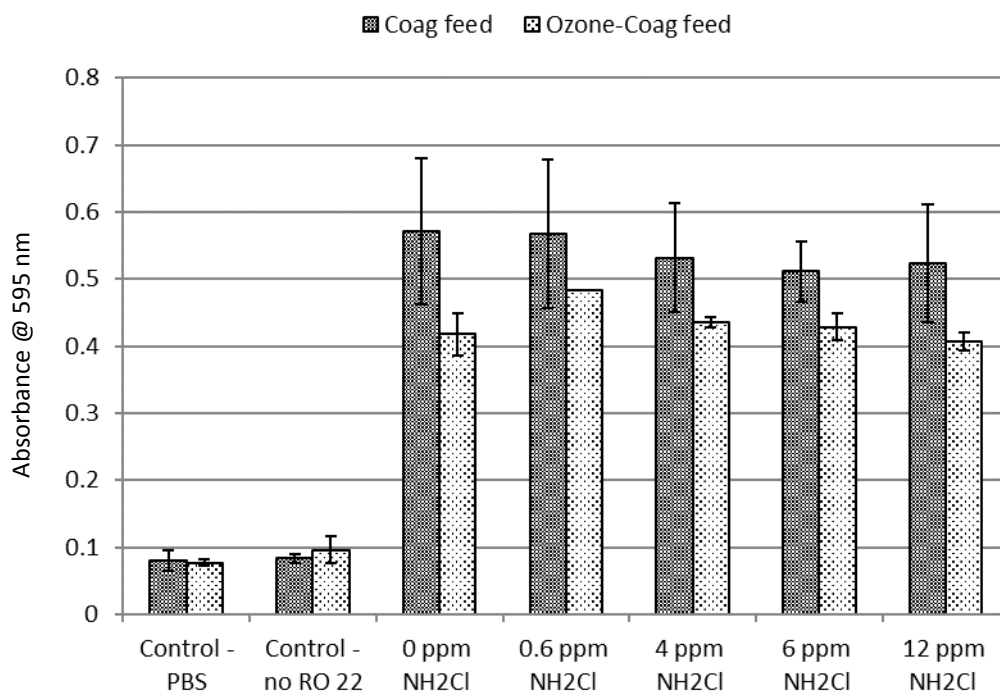
The bioassay analysis showed that the concentration of bacterial cells in the ozone+coagulation+CMF treated water was ~ 20% higher than coagulation+CMF treated water (Figure 6). The results also suggest that bacteria can grow in a liquid medium of ozone+coagulation+CMF treated water, which could be rich in assimilable carbon (and potentially nutrients) compared to coagulation+CMF treated water since more biodegradable organic carbon is present. This confirms the BDOC finding, where more DOC was removed by biological activity. However, improved assimilability of organics does not directly indicate RO biofouling as attachment of cells to the membrane surface and formation of a biofilm, rather than BDOC only, decides whether the water has more or less ability to facilitate biofouling.



**Figure 6:** Absorbance measurement at 570 nm of RO 22 bacteria suspension of each water. Error bars show standard error calculated from the standard deviation.

Figure 7 shows the absorbance of biofilm after removing the bacterial suspension for control, coagulant and ozone-coagulant feeds after 48 h. The preformed chloramine ( $\text{NH}_2\text{Cl}$ ) was dosed at different concentrations to the feed samples with bacteria (RO 22) to observe its effect on inhibiting growth of the biofilm. It was observed that the presence of biofilm growth was slightly less for ozone-coagulation treated water compared to coagulation alone treated water as indicated by lower light absorbances. Interestingly, the biofilm assay showed chloramine had no measurable impact on the biofilm growth control or its removal. However, its known application to control biofouling in RO membranes may work differently to inhibiting biofilm growth which are discussed later during the accelerated RO biofouling tests.

Another interesting feature of the crystal violet assay was the consistently lower biofilm formation when ozone was used. While it was observed that cell growth is enhanced in the presence of water that was treated by ozone (Table 5 and Figure 6), the formation of an actual biofilm which is responsible for biofouling of RO membranes appears suppressed. Biofouling is a complex phenomenon, and recent research has shown that MF pre-treatment of wastewater leads to enhanced biofilm formation [23]. This was found to be due to the removal of ‘antibiofilm’ substances that inhibited growth of the model bacterium *Pseudomonas aeruginosa* PAOI. The MF membrane allowed the passage of lectin-like humics which were able to attach to the RO membrane to form a conditioning layer which in turn facilitated bioadhesion. In our case, it is possible that the lectin-like humic substances were significantly altered by ozone action which reduced their ability to form the essential conditioning layer needed for biofilm establishment. Indeed as shown earlier, ozone action on organics reduced the proportion of humic and hydrophobic substances (Figure 2c) and reduced aromaticity (Table 3). Also, it was noted during analysis of the RO concentrate (Figure 4b), that humic substances can deposit on, or permeate into, the RO membrane which implies they can attach to the polyamide membrane material and further facilitate bioadhesion. The action of ozone to alter their properties limited their ability to be transported through the CMF membrane (Figure 2c), but also apparently improved their ability to be rejected by the RO membrane (Figure 4b).



**Figure 7:** Crystal violet assay measurement at 595 nm for cell density and biofilm production by each water. Error bars show standard error calculated from the standard deviation.

#### *Accelerated RO biofouling test*

RO membranes were tested for accelerated biofouling in a cross flow system of two selected water solutions spiked with RO22 bacteria: coagulation+CMF with chloramine (dosed 2 hours prior to finishing the RO test) and ozone+coagulation+CMF without chloramine dosing. A control with chloramine dosing but no RO22 bacteria was also run. The results of the tests showing the thickness of the fouling layers formed measured by confocal microscopy are presented in Table 6. The addition of RO22 led to additional fouling from biofilm growth on the RO membranes, despite the addition of chloramine. This further supports the crystal violet assay result in Figure 7 where no change in biofilm formation was observed as a result of chloramine dosing. The solution treated with ozone showed slightly thicker fouling layer thickness compared to coagulant only. The thickness of the fouling layer in Table 6 was greater than without ozone, which is the opposite trend to that in Figure 7 which may be due to differences between composite fouling layers and biofilms, as well as effects of permeation and cross flow that occur in the case of RO operation. The differences in the results of the RO biofilm for the crystal violet assay will be now looked at more closely using confocal microscopy to observe the abundance of bacterial cells within the biofilm.






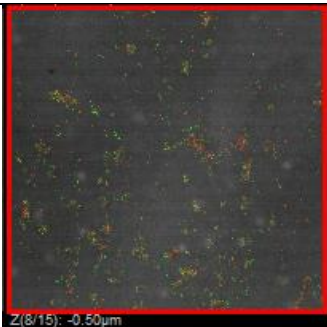
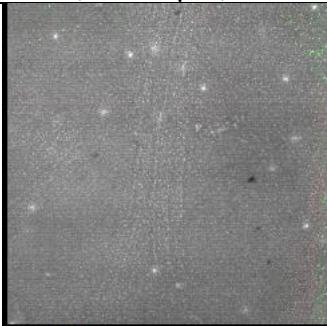
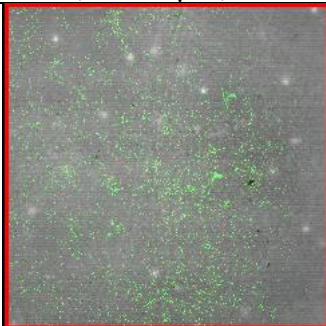
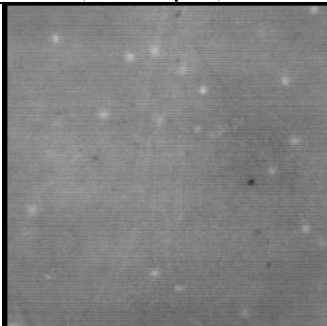
The confocal images presented in Figure 8 show the presence of live (green) and dead (red) cells. In the Figure,  $x$  represents the distance from the RO membrane surface, and  $l$  represents the estimated total fouling layer thickness from Table 6. The ratio  $x/l$  is therefore the relative distance from the RO membrane surface to the fouling layer surface facing the flowing solution. A few live (green) cells appeared on the control membrane without the addition of RO 22, reflecting the presence of low levels of bacteria in the system. In experiments with spiked RO22, many more cells were observed on the membranes. Looking more closely we can see varying quantities and proportions of live and dead cells as a function distance from the membrane surface. For the membrane with coagulant only and chloramine added 2 hours prior to finishing the RO run, live and dead cells were seen near the top of the fouling layer facing the flowing solution at  $x/l = 0.93$  (Supplementary Material Figure S6). Readily available chloramine in the solution was potentially responsible for killing these sessile bacteria. Going deeper to near half the fouling layer thickness ( $x/l = 0.47$ ) in Figure 8 we again see a proportion of both dead (red) and live (green) cells. Closer to the membrane surface ( $x/l = 0.20$  in Figure 8), there were predominantly green (live) cells suggesting that chloramine was not effective for killing these cells, possibly because they were sheltered by the fouling layer above. From the thickness of 4  $\mu\text{m}$  to the membrane surface, cells were predominantly dead (red) (observed at  $x/l = 0.07$  in Figure 8), indicating that the cells that first attached to the membrane surface did not survive during the run, potentially as a result of depletion of nutrients required for cell growth as the water diffuses through the fouling layer. Right at the membrane surface ( $x/l = 0$ ) no cells were found. This could potentially be the conditioning layer, having a thickness of around 2  $\mu\text{m}$ .

In the case of ozone treated water (without chloramine) no cells were seen from the top of the fouling layer facing the flowing solution down to  $x/l = 0.33$  as shown in Figure 8. However reaching  $x/l = 0.27$  saw abundant numbers of predominantly live (green) cells. Like the coagulation pre-treated RO feed, few cells were seen 2  $\mu\text{m}$  from the RO membrane surface ( $x/l = 0.06$ ), but some dead cells appeared at 4.0  $\mu\text{m}$  (Supplementary Material Figure S7). So despite the thicker fouling layer in the presence of ozone treated water as measured by confocal microscopy, it appears the microbial population was much more limited in its thickness compared to the coagulant+CMF pre-treated water case.

**Table 6:** Fouling layer thickness measured after 22 h cross flow RO run with different water samples spiked with RO22 bacteria. Chloramine added 2 hours prior to completion of the RO test (except for ozone-coagulant where no chloramine was dosed).

Water sample	Biofilm thickness, $l$ ( $\mu\text{m}$ )
Coagulant only (control with no RO22 and with 4 mg/L chloramine)	18
Coagulant only (RO22 with 4 mg/L chloramine)	30
Ozone-coagulant (RO22 no chloramine)	36



	Membrane surface	Middle biofilm	Mid - upper biofilm
Control (no RO22 bacteria)	 $x/l = 0.11$ ( $x = 2.0 \mu\text{m}$ )	 $x/l = 0.44$ ( $x = 8.0 \mu\text{m}$ )	 $x/l = 0.61$ ( $x = 11.0 \mu\text{m}$ )
Coag+ CMF feed (with RO22 and chloramin e)	 $x/l = 0.07$ ( $x = 2.0 \mu\text{m}$ )	 $x/l = 0.20$ ( $x = 6.0 \mu\text{m}$ )	 $x/l = 0.47$ ( $x = 14 \mu\text{m}$ )
Ozone+ coag+ CMF feed (with RO22)	 $x/l = 0.06$ ( $x = 2.0 \mu\text{m}$ )	 $x/l = 0.27$ ( $x = 8.0 \mu\text{m}$ )	 $x/l = 0.33$ ( $x = 12.0 \mu\text{m}$ )

**Figure 8:** Confocal images of RO membrane fouled by selected pre-treated wastewaters. The distance from the membrane surface is represented by  $x$ , and shown as the ratio to the estimated total fouling layer thickness  $l$  observed in the confocal imaging.

In considering the application of ozone prior to RO, in a previous pilot trial the lack of biofouling observed was attributed to the use of BAC to digest organics prior to RO [18, 19]. However it may not be required to utilise BAC in all cases. In the 3000 hour pilot trial of RO fed with MBR effluent (one train with added ozone, the other direct from MBR), no operational issues due to biofouling were observed [15]. In fact, they concluded the train with ozone had less biofouling as observed from protein analysis on the lead elements. Therefore, while the biodegradability of organics increases as per the well-known effect of ozone and UVH as shown in Table 5, the prior pilot trial results and our biofouling potential assessment shown in Figure 7 and Figure 8 show that increased biodegradability of organics does not

necessarily lead to increased biofilm formation on RO membranes. On the contrary, both our bioassay result (Figure 7), and the previously reported pilot trial, support the concept that ozone reduces biofouling potential. This could be related to the reduced adhesion of organics on the surface of the membrane that form the conditioning layer needed for a biofilm to commence growth where recent studies have attributed lectin-like humics, which readily pass MF membranes, as being a key compound in forming the conditioning layer leading to RO membrane biofouling [23]. Ozone was shown earlier to generally reduce aromaticity and the hydrophobic proportion of organics, and in particular to reduce the proportion of humic substances in the organics. Removal of humics was even more enhanced by CMF after oxidation (Figure 2) and less likely to deposit or diffuse into the RO membrane (Figure 4b). Further, the oxidised foulants loaded on the RO membranes were more porous (Figure 5). These may have played a key role in limiting the thickness of the active biofilm, despite the organics being more easily assimilable. It appears that ozone (and potentially UVH) treatment prior to RO is not likely to create additional biofouling operation issues. However, further work is needed to confirm similar benefits on other wastewater matrices and process conditions.

The results presented here are representative of a real system, but do not take into account fouling by the actual biomass present in wastewaters, which vary with water source, for example from sea water to wastewater, as well as between lead and tail elements [48]. Further testing is recommended using long term pilot trials on the water to be treated, particularly at recoveries typical of wastewater RO plants (70% to 90%). This would give a more precise determination of actual biofouling risks and location along the membrane using indigenous biological communities.

## Conclusions

In the current study, pre-treatments of RO feed water with coagulation and oxidation processes and filtration with ceramic membranes were proposed for application in wastewater recycling. The main outcomes and recommendations from this work were as follows:

- More sustainable TMP at high CMF flux was achieved when oxidation (either ozone or UVH) and coagulation was applied to the feed water from the WWTP;
- LC-OCD analysis of the various pre-treated waters used as RO feeds showed that CMF can remove the biopolymers and coagulation removes humic substances. Oxidation by ozone or UVH also removed biopolymers, but had a further effect on reducing humic substances concentration. An unconfirmed increase in building blocks and LMW organics was observed in the UVH+coagulation+CMF permeate;
- NDMA analysis of various pre-treated water samples showed an increase in NDMA formation for all samples with chloramine addition, being similar except for UVH which showed a lower relative increase. NDMA increase however was more significant when ozone was used, which could influence the decision for its use (e.g. potable reuse)

in achieving high CMF fluxes. UVH on the other hand showed no NDMA increase, where instead it reduced it to lower than the incoming feed water and may be more favourable in such cases where NDMA must be controlled as it also enables high CMF fluxes;

- The use of ozone or UVH increased the biodegradable organic fraction and growth of RO22 bacteria in the wastewater, but crystal violet assay with RO22 bacteria showed reduced formation of bacterial biofilm communities using ozone+coagulant+CMF pre-treated waters compared to coagulant+CMF pre-treated. Accelerated RO biofouling tests with RO22 bacteria confirmed the findings that despite having a slightly thicker fouling layer, the active bacterial community in the ozone+coagulant+CMF treated water was greatly limited in proximity to the membrane surface compared to coagulant+CMF treated water. This was attributed to the reduction of humic fraction concentrations and alteration of humic chemical properties (including reduced aromaticity), and formation of more porous fouling layers on RO membranes which are less adhesive and more easily removed by flowing water. The findings provide evidence that biofouling due to biofilm formation on RO membranes may not be an issue if upstream oxidation is applied to achieve high CMF fluxes; and
- Chloramine added to biofouling tests did not reduce cell activity in biofilms, but appeared to assist in killing bacteria in the biofilm which extended more into the bulk fluid;

Practical use of CMF as a pre-treatment for RO in advanced water treatment schemes is recommended. However for achieving the high CMF fluxes needed for economical use of ceramic membranes, options must consider the impact of oxidation where ozone leads to potential for NDMA formation while UVH instead could require significant energy. Long term pilot trials in specific contexts are recommended to further explore fouling and operating requirements. As shown in our work with ozone, and as supported by bench and pilot tests by other researchers, oxidation applied upstream of RO leads to minimised organic and biofouling maintenance issues.

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844

845 **List of abbreviations**

846 BAC: biological activated carbon

847 BDOC: biodegradable dissolved organic carbon

848 BSA: bovine serum albumin

849 CMF: ceramic microfiltration

850 DOC: dissolved organic carbon

851 DBP: disinfection by-product

852 HIFI: hydraulic irreversible fouling index

853 LC-OCD: liquid size exclusion chromatography with organic carbon detection

854 LMW: low molecular weight

855 MF: microfiltration

856 MBR: membrane bioreactor

857 NDMA: N-nitrosodimethylamine

858 PACl: polyaluminium chloride

859 PS: polysulfone

860 PVDF: polyvinylidene fluoride

861 RO: reverse osmosis

862 SEM: scanning electron microscopy

863 SRP: salt reduction plant

864 SUVA: specific UV absorbance

865 TDS: total dissolved solids

866 TMP: transmembrane pressure

867 TOrC: trace organic compounds

868 TSB: tryptone soy broth

869 UF: ultrafiltration

870 UMFI: unified membrane fouling index

871 UVH: ultraviolet/hydrogen peroxide

872 WWTP: wastewater treatment plant

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## 874 References

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876 [1] S.G. Lehman and L. Liu, *Application of ceramic membranes with pre-ozonation for treatment of*  
877 *secondary wastewater effluent*. Water Research, 2009. **43**(7): p. 2020-2028.

878 [2] N. Dow, D. Murphy, J. Clement, and M. Duke, *Outcomes of the Australian Ozone/Ceramic*  
879 *Membrane Trial on Secondary Effluent*. AWA Water, 2013. **40**(6): p. 45-51.

880 [3] N. Dow, J. Roehr, D. Murphy, L. Solomon, J. Mieog, J. Blackbeard, S. Gray, N. Milne, B. Zhu, A.  
881 Gooding, J. Currie, G. Roeszler, J. Clement, and M. Duke, *Fouling mechanisms and reduced*  
882 *chemical potential of ceramic membranes combined with ozone*. Water Practice &  
883 Technology, 2015. **10**(4): p. 806-813.

884 [4] J. Haberkamp, A.S. Ruhl, M. Ernst, and M. Jekel, *Impact of coagulation and adsorption on DOC*  
885 *fractions of secondary effluent and resulting fouling behaviour in ultrafiltration*. Water  
886 Research, 2007. **41**(17): p. 3794-3802.

887 [5] W.S. Guo, S. Vigneswaran, and H.H. Ngo, *Effect of flocculation and/or adsorption as pretreatment*  
888 *on the critical flux of crossflow microfiltration*. Desalination, 2005. **172**(1): p. 53-62.

889 [6] E. Vasyukova, R. Proft, J. Jousten, I. Slavik, and W. Uhl, *Removal of natural organic matter and*  
890 *trihalomethane formation potential in a full-scale drinking water treatment plant*. Water  
891 Science and Technology: Water Supply, 2013. **13**(4): p. 1099-1108.

892 [7] L. Fan, T. Nguyen, F.A. Roddick, and J.L. Harris, *Low-pressure membrane filtration of secondary*  
893 *effluent in water reuse: Pre-treatment for fouling reduction*. Journal of Membrane Science,  
894 2008. **320**(1-2): p. 135-142.

895 [8] O. González, A. Justo, J. Bacardit, E. Ferrero, J.J. Malfeito, and C. Sans, *Characterization and fate of*  
896 *effluent organic matter treated with UV/H<sub>2</sub>O<sub>2</sub> and ozonation*. Chemical Engineering Journal,  
897 2013. **226**(0): p. 402-408.

898 [9] W. Yu, D. Zhang, and N.J.D. Graham, *Membrane fouling by extracellular polymeric substances after*  
899 *ozone pre-treatment: Variation of nano-particles size*. Water Research, 2017. **120**: p. 146-155.

900 [10] S. Lee, K. Lee, W.M. Wan, and Y. Choi, *Comparison of membrane permeability and a fouling*  
901 *mechanism by pre-ozonation followed by membrane filtration and residual ozone in*  
902 *membrane cells*. Desalination, 2005. **178**(1): p. 287-294.

903 [11] M. Hashino, Y. Mori, Y. Fujii, N. Motoyama, N. Kadokawa, H. Hoshikawa, W. Nishijima, and M.  
904 Okada, *Pilot plant evaluation of an ozone-microfiltration system for drinking water treatment*.  
905 Water Science and Technology, 2000. **41**(10-11): p. 17-23.

906 [12] Y.G. Park, *Effect of ozonation for reducing membrane-fouling in the UF membrane*. Desalination,  
907 2002. **147**(1): p. 43-48.

908 [13] X. Wang, L. Wang, Y. Liu, and W. Duan, *Ozonation pretreatment for ultrafiltration of the secondary*  
909 *effluent*. Journal of Membrane Science, 2007. **287**(2): p. 187-191.

910 [14] B. Zhu, Y. Hu, S. Kennedy, N. Milne, G. Morris, W. Jin, S. Gray, and M. Duke, *Dual function filtration*  
911 *and catalytic breakdown of organic pollutants in wastewater using ozonation with titania and*  
912 *alumina membranes*. Journal of Membrane Science, 2011. **378**(1-2): p. 61-72.

913 [15] B.D. Stanford, A.N. Pisarenko, S.A. Snyder, and R.D. Holbrook, *Pilot-scale oxidative technologies*  
914 *for reducing fouling potential in water reuse and drinking water membranes*. 2013, Water  
915 Reuse Association.

916 [16] B.D. Stanford, A.N. Pisarenko, R.D. Holbrook, and S.A. Snyder, *Preozonation effects on the*  
917 *reduction of Reverse Osmosis Membrane Fouling in Water Reuse*. Ozone: Science &  
918 Engineering, 2011. **33**(5): p. 379-388.

- [17] J. Zhang, K. Northcott, M. Duke, P. Scales, and S.R. Gray, *Influence of pre-treatment combinations on RO membrane fouling*. Desalination, 2016. **393**: p. 120-126.
- [18] J. Zhang, A. Knight, M. Duke, K. Northcott, M. Packer, P.J. Scales, and S.R. Gray, *A new integrated potable reuse process for a small remote community in Antarctica*. Process Safety and Environmental Protection, 2016. **104, Part A**: p. 196-208.
- [19] J. Zhang, M. Duke, K. Northcott, M. Packer, M. Allinson, G. Allinson, K. Kadokami, J. Tan, S. Allard, J.-P. Croué, A. Knight, P. Scales, and S. Gray, *Small scale direct Potable Reuse (DPR) Project for a Remote Area*. Water, 2017. **9**(2): p. 94.
- [20] B.S. Oh, H.Y. Jang, Y.J. Jung, and J.-W. Kang, *Microfiltration of MS2 bacteriophage: Effect of ozone on membrane fouling*. Journal of Membrane Science, 2007. **306**(1–2): p. 244-252.
- [21] S.T. Nguyen and F.A. Roddick, *Effects of ozonation and biological activated carbon filtration on membrane fouling in ultrafiltration of an activated sludge effluent*. Journal of Membrane Science, 2010. **363**(1–2): p. 271-277.
- [22] K.I. Abdul Hamid, P. Sanciolo, S. Gray, M. Duke, and S. Muthukumaran, *Impact of ozonation and biological activated carbon filtration on ceramic membrane fouling*. Water Research, 2017.
- [23] H. Winters, T.H. Chong, A.G. Fane, W. Krantz, M. Rzechowicz, and N. Saeidi, *The involvement of lectins and lectin-like humic substances in biofilm formation on RO membranes - is TEP important?* Desalination, 2016. **399**: p. 61-68.
- [24] W.A. Mitch, J.O. Sharp, R.R. Trussell, R.L. Valentine, L. Alvarez-Cohen, and D.L. Sedlak, *N-Nitrosodimethylamine (NDMA) as a drinking water contaminant: a review*. Environmental Engineering Science, 2004. **20**(5): p. 389-404.
- [25] P. Andrzejewski, B. Kasprzyk-Hordern, and J. Nawrocki, *N-nitrosodimethylamine (NDMA) formation during ozonation of dimethylamine-containing waters*. Water Research, 2008. **42**(4–5): p. 863-870.
- [26] P. Andrzejewski and J. Nawrocki, *N-nitrosodimethylamine (NDMA) as a product of potassium permanganate reaction with aqueous solutions of dimethylamine (DMA)*. Water Research, 2009. **43**(5): p. 1219-1228.
- [27] T. Bond, J. Huang, M.R. Templeton, and N. Graham, *Occurrence and control of nitrogenous disinfection by-products in drinking water – A review*. Water Research, 2011. **45**(15): p. 4341-4354.
- [28] C.K. Schmidt and H.-J. Brauch, *N,N-Dimethylsulfamide as precursor for N-Nitrosodimethylamine (NDMA) formation upon Ozonation and its Fate During Drinking Water Treatment*. Environmental Science & Technology, 2008. **42**(17): p. 6340-6346.
- [29] APHA, AWWA, and WEF, *Standard methods for the examination of water & wastewater*. 21st ed, ed. A. Eaton, L.S. Clesceri, A.E. Greenberg, and E.W. Rice. 2005: American Water Works Association (AWWA)
- [30] H. Bader, V. Sturzenegger, and J. Hoigné, *Photometric method for the determination of low concentrations of hydrogen peroxide by the peroxidase catalyzed oxidation of N,N-diethyl-p-phenylenediamine (DPD)*. Water Research, 1988. **22**(9): p. 1109-1115.
- [31] D.T. Myat, M. Mergen, O. Zhao, M.B. Stewart, J.D. Orbell, and S. Gray, *Characterisation of organic matter in IX and PACl treated wastewater in relation to the fouling of a hydrophobic polypropylene membrane*. Water Research, 2012. **46**(16): p. 5151-5164.
- [32] H. Huang, T.A. Young, and J.G. Jacangelo, *Unified membrane fouling index for low Pressure Membrane Filtration of Natural Waters: Principles and Methodology*. Environmental Science & Technology, 2008. **42**(3): p. 714-720.
- [33] A.H. Nguyen, J.E. Tobiasson, and K.J. Howe, *Fouling indices for low pressure hollow fiber membrane performance assessment*. Water Research, 2011. **45**(8): p. 2627-2637.
- [34] A.N. Pisarenko, D. Yan, S.A. Snyder, and B.D. Stanford, *Comparing oxidative organic oouling Control in RO Membrane Applications*. IDA Journal of Desalination and Water Reuse, 2011. **3**(2): p. 45-49.

- [35] S.A. Huber, A. Balz, M. Abert, and W. Pronk, *Characterisation of aquatic humic and non-humic matter with size-exclusion chromatography – organic carbon detection – organic nitrogen detection (LC-OCD-OND)*. Water Research, 2011. **45**(2): p. 879-885.
- [36] S.A. Huber and F.H. Frimmel, *A new method for the characterization of Organic Carbon in Aquatic Systems*. International Journal of Environmental Analytical Chemistry, 1992. **49**(1-2): p. 49-57.
- [37] J.C. Joret and Y. Levi, *Methode rapide d'évaluation du carbone éliminable des eaux par voie biologique*. Trib. Cebedeau, 1986. **510**: p. 3-9.
- [38] C. Volk, C. Renner, P. Roche, H. Paillard, and J.C. Joret, *Effects of ozone on the production Of Biodegradable Dissolved Organic Carbon (BDOC) During Water Treatment*. Ozone: Science & Engineering, 1993. **15**(5): p. 389-404.
- [39] J.W.A. Charrois, M.W. Arend, K.L. Froese, and S.E. Hrudey, *Detecting N-nitrosamines in drinking water at Nanogram per Liter Levels Using Ammonia Positive Chemical Ionization*. Environmental Science & Technology, 2004. **38**(18): p. 4835-4841.
- [40] G.A. O'Toole, *Microtiter dish biofilm formation assay*. Journal of Visualized Experiments : JoVE, 2011(47): p. 2437.
- [41] V. Nagaraj, L. Skillman, D. Li, A. Foreman, Z. Xie, and G. Ho, *Characterisation of extracellular polysaccharides from bacteria isolated from a full-scale desalination plant*. Desalination, 2017. **418**: p. 9-18.
- [42] M.R. Jekel, *Flocculation effects of ozone*. Ozone: Science & Engineering, 1994. **16**(1): p. 55-66.
- [43] B. Domenjoud, N. Cortés-Francisco, R. Guastalli Andrea, J. Caixach, S. Esplugas, and S. Baig, *Ozonation of municipal secondary ffluent; Removal of Hazardous Micropollutants and Related Changes of Organic Matter Composition*, in *Journal of Advanced Oxidation Technologies*. 2011. p. 138.
- [44] R.H.S. Jansen, A. Zwijnenburg, W.G.J. van der Meer, and M. Wessling, *Outside-in trimming of humic Substances During Ozonation in a Membrane Contactor*. Environmental Science & Technology, 2006. **40**(20): p. 6460-6465.
- [45] X. Liao, C. Chen, B. Yuan, J. Wang, and X. Zhang, *Control of nitrosamines, THMs, and HAAs in heavily impacted Water With O3-BAC*. Journal - American Water Works Association, 2017. **109**(6): p. E215-E225.
- [46] D. Gerrity, A.N. Pisarenko, E. Marti, R.A. Trenholm, F. Gerringer, J. Reungoat, and E. Dickenson, *Nitrosamines in pilot-scale and full-scale wastewater treatment plants with ozonation*. Water Research, 2015. **72**: p. 251-261.
- [47] S.W. Krasner, W.A. Mitch, D.L. McCurry, D. Hanigan, and P. Westerhoff, *Formation, precursors, control, and occurrence of nitrosamines in drinking water: A review*. Water Research, 2013. **47**(13): p. 4433-4450.
- [48] M.T. Khan, M. Busch, V.G. Molina, A.-H. Emwas, C. Aubry, and J.-P. Croue, *How different is the composition of the fouling layer of wastewater reuse and seawater desalination RO membranes?* Water Research, 2014. **59**: p. 271-282.
- [49] J.S. Vrouwenvelder and D. van der Kooij, *Diagnosis, prediction and prevention of biofouling of NF and RO membranes*. Desalination, 2001. **139**(1): p. 65-71.